Serial No. 10/089.776 Docket No. 29288 5600

## Amendments To Claims:

This listing of claims will replace all prior versions, and listings, of claims in the application:

## Listing of Claims:

(Currently Amended) A time-resolved fluoroimmunoassay (TR-FIA) method for detecting a cytokine in a biological fluid sample, comprising:

subjecting the biological fluid sample to a heat treatment under non-denaturing temperature conditions of about 37°C to about 55°C for the cytokine; and

forming a composite by (a) binding a first antibody, including a portion bound to a solid phase and a region bindable to a cytokine, to a solid phase; (b) adding the sample containing the cytokine; (c) binding a second antibody, including a region bindable to the cytokine and a portion to which biotin is bound, to the cytokine; (d) adding a conjugate including streptoavidin or avidin and a fluorescent structural portion capable of being complexed with a lanthanoid metal ion; and (e) adding a lanthanoid metal ion to bind the conjugate of (d), the composite being formed on the solid phase; and

measuring fluorescence of the fluorescent structural portion which has been complexed with the lanthanoid metal ion,

wherein the method comprises a step of washing after each of steps (a) to (c); and

wherein the cytokine is a cytokine belonging to the chemokine family, and wherein the fluorescent structural portion is represented by General Formula (I): **(I)** 

(where R is a residue which is a functional group capable of forming a covalent bond with a protein; Ar is a hydrocarbon group having a conjugated double bond system; n is an integer equal to or greater than 1; and X is a fluorine atom or a group represented by General Formula (II):

$$-C(=0)-CH_2-C(=0)-Ar-R$$
 (II).

(Original) A method according to claim 1, wherein the lanthanoid 1 metal 2. ion is europium.

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(Previously Presented) A method according to claim 1, wherein the 3. fluorescent structural portion is represented by General Formula (III):

$$R-Ar-(C(=0)-CH_2-C(=0)-C_nF_{2n})_2$$
 (III)

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(where R. Ar. and n have the same definitions as in claim 1).

 (Original) A method according to claim 3, wherein the fluorescent structural portion is 4,4-bis(1", 1", 1", 2", 2", 3", 3"heptafluoro-4", 6"-hexanedion-6"-yl)sulpho-oterphenyl.

- (Original) A method according to claim 1, wherein 10 to 60 units of the fluorescent structural portion are present per molecule of streptoavidin or avidin in the conjugate.
- (Original) A method according to claim 1, wherein the step of measuring fluorescence is performed without allowing the composite formed on the solid phase to dissociate.
- (Original) A method according to claim 1, wherein the step of measuring fluorescence is performed after allowing the composite formed on the solid phase to dissociate.
  - 8. (Cancelled)
- (Original) A method according to claim 1, wherein the cytokine is a CXC chemokine.
- (Original) A method according to claim 9, wherein the cytokine is stromal cell-derived factor-1 (SDF-1).
- (Original) A method according to claim 1, wherein the biological fluid sample is plasma or whole blood.
- (Original) A method according to claim 1, further comprising, before the step of forming the composite, a step of diluting the biological fluid sample with a buffer solution used for sample dilution,

wherein the buffer solution used for sample dilution is 0.01 to 0.1 M trishydrochloric acid whose pH is 7.3 to about 8.3, the buffer solution containing 0.1 to 0.3% of bovine serum albumin, 0.05 to 0.2% of sodium azide, and 0.5 to 1.5% of sodium chloride.

- 13. (Cancelled)
- (Original) A method according to claim 1, further comprising, before the step of measuring fluorescence, a step of washing the composite formed on the solid phase with a buffer solution used for washing,

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wherein the buffer solution used for washing the composite is 0.01 to 0.1 M trishydrochloric acid whose pH is 8.5 to about 9.5, the buffer solution containing 0.01 to 0.1% polyoxyethylenesorbitan monolaurate.

- 15. (Original) A method according to claim 1, wherein the solid phase is a microtiter plate having an IgG adsorption ability of 50 to 200 ng/cm<sup>2</sup>.
- 16. (Currently Amended) A kit for a time-resolved fluoroimmunoassay (TR-FIA) method for detecting a cytokine in a biological fluid sample, which has been subjected to heat treatment under non-denaturing temperature conditions for the cytokine of about 37°C-te-about 55°C, comprising: a first antibody including a portion bound to a solid phase and a region bindable to a cytokine; a second antibody including a region bindable to the cytokine and a portion to which biotin is bound; a conjugate including streptoavidin or avidin and a fluorescent structural portion capable of being complexed with a lanthanoid metal ion; and the lanthanoid metal ion,

wherein the cytokine is a cytokine belonging to the chemokine family, and wherein the fluorescent structural portion is represented by General Formula (I):  $R-Ar-C(=0)-CH_2-C(=0)-C_2F_{2n}-X$  (1)

(where R is a residue which is a functional group capable of forming a covalent bond with a protein; Ar is a hydrocarbon group having a conjugated double bond system; n is an integer equal to or greater than 1; and X is a fluorine atom or a group represented by General Formula (II):

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$$-C(=0)-CH_2-C(=0)-Ar-R$$
 (II).

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